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Workshop report

Nonclinical safety testing of biopharmaceuticals – Addressing current challenges of these novel and emerging therapies

Frank R. Brennan [a, *], Andreas Baumann [b], Guenter Blaich [c], Lolke de Haan [d], Rajni Fagg [e], Andrea Kiessling [f], Sven Kronenberg [g], Mathias Locher [h], Mark Milton [i], Jay Tibbitts [a], Peter Ulrich [f], Lucinda Weir [e]

[a] UCB Pharma, Slough, UK
[b] Bayer Pharma AG, Berlin, Germany
[c] Abbvie GmbH, Ludwigshafen, Germany
[d] Medimmune LLC, Cambridge, UK
[e] Glaxo Smithkline Pharma, Stevenage, UK
[f] Novartis Pharma, Basel, Switzerland
[g] Roche Pharmaceutical Research and Early Development, Pharmaceutical Sciences, Roche Innovation Center Basel, Switzerland
[h] Covagen, Zurich, Switzerland
[i] Novartis Pharma, Boston, USA

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ABSTRACT

Non-clinical safety testing of biopharmaceuticals can present significant challenges to human risk assessment with these often innovative and complex drugs. Hot Topics in this field were discussed recently at the 4th Annual European Biosafe General Membership meeting. In this feature article, the presentations and subsequent discussions from the main sessions are summarized. The topics covered include: (i) wanted versus unwanted immune activation, (ii) bi-specific protein scaffolds, (iii) use of Pharmacokinetic (PK)/Pharmacodynamic (PD) data to impact/optimize toxicology study design, (iv) cytokine release and challenges to human translation (v) safety testing of cell and gene therapies including chimeric antigen receptor T (CAR-T) cells and retroviral vectors and (vi) biopharmaceutical development strategies encompassing a range of diverse topics including optimizing entry of monoclonal antibodies (mAbs) into the brain, safety testing of therapeutic vaccines, non-clinical testing of biosimilars, infection in toxicology studies with immunomodulators and challenges to human risk assessment, maternal and infant anti-drug antibody (ADA) development and impact in non-human primate (NHP) developmental toxicity studies, and a summary of an NC3Rs workshop on the future vision for non-clinical safety assessment of biopharmaceuticals.

1. Introduction

Biosafe is the Preclinical Safety expert group of the Biotechnology Industry Organization (BIO), which has been tasked with the mission to serve as a resource for BIO members and BIO staff by identifying and responding to key scientific and regulatory issues related to the preclinical safety evaluation of biopharmaceutical products. Meetings of the Biosafe General Membership are held both in the US and in Europe on an annual basis (Kronenberg et al., 2013, Baumann et al., 2014). The 4th Annual Biosafe European General Membership meeting was hosted by UCB Pharma on November 5–6, 2014 in Windsor, UK. The 140 scientists, predominantly from Europe but also from the US, with pharmacology, toxicology, pathology, PK or bioanalytical backgrounds, represented global big and mid-size pharmaceutical companies, small biotechnology companies and contract research organizations (CROs) including GSK, Medimmune, Genentech, Abbvie, UCB, Roche, Novartis, Pfizer, Bayer, Bristol Myers Squibb, Covagen, Covance and many more. Attendees shared experiences and insights into the nonclinical safety assessment of biopharmaceuticals, including mAbs, recombinant proteins, cell and gene therapies and vaccines. The meeting covered several nonclinical safety issues,

* Corresponding author. UCB Pharma, 208 Bath Road, Slough SL1 3WE, UK.
E-mail address: frank.brennan@ucb.com (F.R. Brennan).

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with a strong focus on immunomodulation, covering topics such as adverse immune-mediated toxicities (due to ADA, cell activation and cytokine release etc.) and the challenges to human translation, PK/PD-driven toxicity study design, non-clinical testing of bi-specifics (where novel biology can complicate human dose selection and safety assessment), and the challenges in identifying relevant models for safety testing of gene and cell therapies. During each session, case studies were presented and followed by podium discussions, and in some cases, round table discussions.

2. Wanted vs unwanted immune activation

In the first session of the meeting, chaired by **Sven Kronenberg (Roche)** and **Lolke de Haan (Medimmune)**, the primary focus was on assessment of nonclinical safety of novel immune-mediated therapies in oncology and ADA-mediated toxicity. Anti-cancer immune-mediated therapies include (i) immunomodulatory agents, such as checkpoint inhibitors, adjuvants, cytokines, costimulatory molecules, (ii) immune cell recruiting agents, such as Bispecific T-cell Engagers (BiTE®s) and Immune-Mobilizing Monoclonal T-cell receptors Against Cancer (ImmTACs), and (iii) other therapies, including vaccines, cell therapies, modalities modulating the tumor microenvironment etc. (see Fig. 1). From a pharmacology perspective, efficacy with these modalities is often difficult to demonstrate as the traditionally used mouse xenograft models can often not be used, because few of these modalities are rodent cross-reactive, and these therapies often require a fully functional immune system for activity. Moreover, such molecules are often very immunogenic in mice. From a nonclinical safety perspective, there are clear limitations in the predictivity of the traditional nonclinical safety testing models with respect to assessment of immune-mediated adverse effects (e.g. due to limited target expression in naive, healthy animals that are poor predictors for anti-tumor effects and human immunogenicity). The immune-stimulatory nature of these modalities may lead to enhancement of ADA responses and compromise the validity of the toxicity study or lead to ADA-mediated toxicity. Finally, as a consequence of the fact that most modalities are cross-reactive to the NHP only, assessment of the activity of the drug candidate is often limited to demonstration of target engagement ex vivo (e.g. receptor occupancy assays, suppression of soluble target etc) and no clear pharmacology can be demonstrated.

Against this background, **Rod Prell (Genentech)** presented a case study on MPDL3280A, an engineered humanized IgG1 mAb directed against PD-L1. Both the mouse and cynomolgus monkey are pharmacologically-relevant species for MPDL3280A. In a 15-day pilot study in the mouse, MPDL3280A induced neuropathy, characterized by minimal axonal degeneration with lymphocytic infiltration. Due to induction of high levels of circulating ADAs, repeat-dose toxicity studies of longer duration were not feasible. In an 8-week study in cynomolgus monkeys, exposure to MPDL3280A could be maintained despite high levels of ADAs, and periarteritis/arteritis was noted, characterized as mixed inflammation around and involving blood vessels and medium-sized muscular arteries. There were no clinical signs associated with the microscopic lesions in either species. The observed neuropathy and arteritis were considered consistent with the anticipated mode of action of MPDL3280A, and underscored the hypothetical concerns around

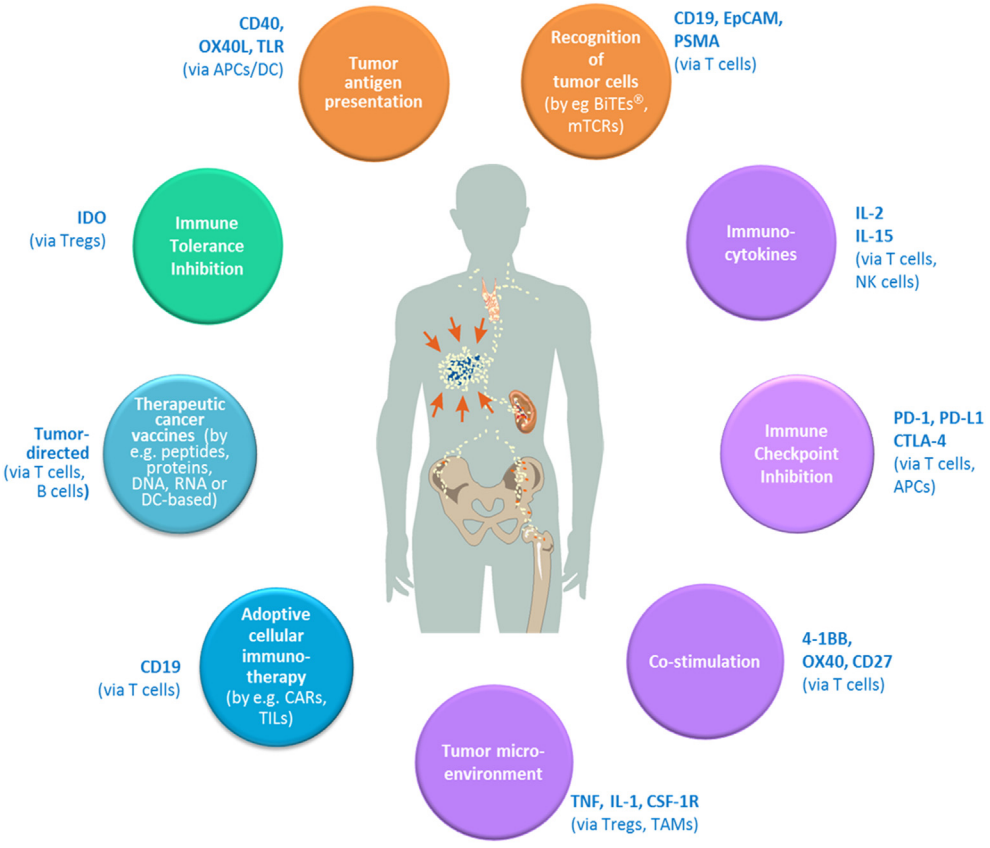

Fig. 1. Cancer immunotherapy (CIT) – an evolving field of multiple immunomodulatory targets, and platforms. Circles represent different research areas/modalities in CIT, blue text indicates examples of published targets and involved immune cells. APC: Antigen-presenting Cells, BiTE®: Bispecific T cell Engager; CAR: Chimeric Antigen Receptor; DC: Dendritic Cells; mTCR: monoclonal T cell Receptor; NK cells: Natural killer cells; TAM: Tumor-associated Macrophages; TIL: Tumor-infiltrating Lymphocytes. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)